

Amendments to the Claims

1. (Currently amended) A method for adjusting the yield and purity of a proteinase inhibitor isolated from tissue of a plant, comprising the steps of:
 - (a) extracting the protease inhibitor and other protein products from the plant tissue by preparing a mixture of an alcohol-free solvent, an organic acid, and comminuted plant tissue to form a solid fraction and a liquid fraction comprising the protease inhibitor and other protein products;
 - (b) heating the liquid fraction to a temperature and for a time period sufficient to denature at least some of the other protein products without substantially denaturing the protease inhibitor;
 - (c) adjusting the temperature and time period of the heat treatment step to selectively affect the purity and yield of the protease inhibitor; and
 - (d) removing the denatured protein products to prepare a clarified extract solution.
2. (Original) The method of claim 1 wherein the solvent comprises formic acid and sodium chloride.
3. (Original) The method of claim 2 wherein the solvent comprises about 0.5% to about 2.5 % formic acid and 0 to 3.0 N sodium chloride.
4. (Previously amended) The method of claim 1 wherein the heating step is conducted at between about 60° to about 90° C.
5. (Previously amended) The method of claim 4 wherein the heating step is conducted for between about 30 to about 180 minutes.
6. (Previously amended) The method of claim 4 wherein the heating step is conducted at a temperature greater than about 75° C to increase the purity of the protease inhibitor.

7. (Original) The method of claim 4 wherein yield of the protease inhibitor is increased by selecting a temperature less than about 75° C.
8. (Currently cancelled) The method of claim 1 wherein the adjusting step is conducted to increase the temperature of the heat treatment step and simultaneously to decrease the duration of the heat treatment step.
9. (Currently cancelled) The method of claim 1 wherein the adjusting step is conducted to decrease the temperature of the heat treatment step and simultaneously increase the duration of the heat treatment step.
10. (Original) The method of claim 1 wherein the step of removing the denatured proteins is carried out by centrifugation.
11. (Previously amended) The method of claim 1, further comprising filtering the clarified extract to remove impurities having a molecular weight below that of the proteinase inhibitor.
12. (Previously amended) The method of claim 11 wherein filtration is conducted on an ultrafiltration membrane having a molecular weight cut-off rating of about 5 KD to about 10 KD.
13. (Previously amended) The method of claim 11 wherein a buffer solution comprising an aqueous solution of ammonium bicarbonate is added during filtration.
14. (Original) The method of claim 13 wherein the buffer is between about 50 and about 500 mM ammonium bicarbonate.
15. (Previously amended) The method of claim 11 wherein the clarified extract is concentrated to less than one-fifth of the starting volume during filtration.
16. (Previously amended) The method of claim 15 wherein the filtration step further comprises washing the clarified extract with up to ten volumes of filtration buffer.

17. (New) A method for adjusting the yield and purity of a proteinase inhibitor isolated from tissue of a plant, comprising the steps of:

- (a) extracting the protease inhibitor and other protein products from the plant tissue by preparing a mixture of an alcohol-free solvent, an organic acid, and comminuted plant tissue to form a solid fraction and a liquid fraction comprising the protease inhibitor and other protein products;
- (b) heating the liquid fraction to a temperature and for a time period sufficient to denature at least some of the other protein products without substantially denaturing the protease inhibitor;
- (c) increasing the temperature of the heat treatment step and simultaneously decreasing the duration of the heat treatment step to selectively affect the purity and yield of the protease inhibitor; and
- (d) removing the denatured protein products to prepare a clarified extract solution.

18. (New) A method for adjusting the yield and purity of a proteinase inhibitor isolated from tissue of a plant, comprising the steps of:

- (a) extracting the protease inhibitor and other protein products from the plant tissue by preparing a mixture of an alcohol-free solvent, an organic acid, and comminuted plant tissue to form a solid fraction and a liquid fraction comprising the protease inhibitor and other protein products;
- (b) heating the liquid fraction to a temperature and for a time period sufficient to denature at least some of the other protein products without substantially denaturing the protease inhibitor;
- (c) decreasing the temperature of the heat treatment step and simultaneously increasing the duration of the heat treatment step to selectively affect the purity and yield of the protease inhibitor; and
- (d) removing the denatured protein products to prepare a clarified extract solution.